No attempt should be made to submit such cases to medical treatment, for no one can foretell when an acute perforation will supervene; also where communication has been established with neighboring hollow viscera.

3. In all chronically recurring bleeding ulcers.
4. In hour-glass contractions where there is a tendency to narrowing of the connecting isthmus.
5. Those cases with frequent recurrence of symptoms in which repeated dietetic courses are required which admit the failure of controlling the symptoms by medical means.
6. Too long a time must not be spent with intense medical treatment if symptoms do not abate.

**Summary.** 1. A series of 14 cases of gastric ulcer is reported in which complete healing of the ulcer was obtained by medical treatment and has been maintained for periods up to four and a half years.

2. The question whether these ulcers have been permanently cured is discussed and their possible relationship to constitutional states in regard to etiology and recurrence pointed out.

3. The conditions which determine whether medical treatment is to be replaced by surgical interference are described.

---

# A MICRO-METHOD FOR THE DETERMINATION OF SUGAR IN SMALL AMOUNTS OF BLOOD.

By H. O. Pollock, M.D.,

AND

W. S. McEllroy, M.D.,

PITTSBURGH, PA.

(From the Chemical Laboratories, School of Medicine, University of Pittsburgh and Mercy Hospital.)

Blood-sugar determinations are of practical value in the diagnosis and control of certain diseases of metabolism. As indicated by Allen[1] and Williams[2] this applies particularly to diabetes.

The value of blood-sugar determinations, however, is largely dependent upon their degree of accuracy, for faulty determinations are worse than useless, as they may lead to erroneous conclusions. Many methods and modifications have been introduced. In this country the colorimetric methods of Lewis and Benedict[3] and of

---

[1] Total Dietary Regulation in the Treatment of Diabetes, Monograph No. 11, Rockefeller Institute for Medical Research, 1919.

[2] The Clinical Significance of Blood Sugar in Diabetes Mellitus, Arch. Int. Med., 1919, 23, 546.

[3] A Modification of the Lewis-Benedict Method for the Determination of Sugar in the Blood, Jour. Biol. Chem., 1918, 34, 203.

Folin and Wu[4] are largely employed. Although only a small quantity of blood (usually 2 cc or more) is required for these methods it is necessary to obtain it from a vein.

There is no serious objection to venous puncture ordinarily, but at times it is difficult or undesirable, particularly when determinations are made on children or repeated determinations on adults.

To overcome the necessity for venous puncture, attempts have been made to adapt many of the blood-sugar methods to smaller volumes of blood which can be drawn from the finger tip or lobe of the ear. From the standpoint of accuracy such micro-modifications have not been found very satisfactory.

The method to be described is an adaptation of that of Folin and Wu to 0.2 cc of blood drawn from the finger or lobe of the ear by simple puncture. We have found it of value in supplementing the original method in cases in which it is desirable to avoid repeated venous puncture, and have obtained accurate and reliable results as judged by control determinations by the Folin-Wu method. One advantage of the method is that the final colorimetric comparison can be made in any accurate type of colorimeter.

**Solutions.** No change has been made in the reagents used in the original method of Folin and Wu, and this obviates the necessity of preparing a separate set of solutions. The directions for their preparation, taken from the paper of Folin and Wu, are included here merely as a matter of convenience.

1. *Alkaline Copper Solution.* Dissolve 40 gm. of pure anhydrous sodium carbonate in about 400 cc of water and transfer to a liter flask. Add 7.5 gm. of tartaric acid, and when the latter has dissolved add 4.5 gm. of crystallized copper sulphate. Mix and make up to a volume of 1 liter. If the chemicals used are not pure a sediment of cuprous oxide may form in the course of one or two weeks. If this should happen, remove the clear supernatant reagent with a siphon or filter through a good quality filter paper. The reagent seems to keep indefinitely. To test for absence of cuprous copper in the solution, transfer 2 cc to a test tube and add 2 cc of the molybdate phosphate solution; the deep blue color of the copper should almost completely vanish.

2. *Molybdate Phosphate Solution.* Transfer to a liter beaker 35 gm. of molybdic acid and 5 gm. of sodium tungstate. Add 200 cc of 10 per cent sodium hydroxide and 200 cc of water. Boil vigorously for twenty to forty minutes so as to remove nearly the whole of the ammonia present in the molybdic acid. Cool, dilute to about 350 cc and add 125 cc of concentrated (85 per cent) phosphoric acid. Dilute to 500 cc.

3. *A 10 per cent solution of sodium tungstate.*

---

[4] A System of Blood Analysis. Supplement I. A Simplified and Improved Method for the Determination of Sugar, Jour. Biol. Chem., 1920, 41, 367.

4. *A two-thirds normal solution of sulphuric acid.*

5. *Standard Sugar Solutions.* Three standard sugar solutions should be on hand.

(*a*) A stock solution, 1 per cent dextrose or invert sugar, preserved with xylene or toluene. Dissolve 1 gm. of pure anhydrous dextrose in water and dilute to a volume of 100 cc. Mix, add a few drops of toluene or xylene and bottle. If pure dextrose is not available a standard solution of invert sugar made from cane sugar is equally useful. Transfer exactly 1 gm. of cane sugar to a 100 cc volumetric flask; add 20 cc of normal hydrochloric acid and let the mixture stand overnight at room temperature (or rotate the flask and contents continuously for ten minutes in a water-bath at 70° C.). Add 1.68 gm. of sodium bicarbonate and about 0.2 gm. of sodium acetate to neutralize the hydrochloric acid. Shake a few minutes to remove most of the carbonic acid and fill to the 100 cc mark with water. Then add 5 cc more of water (1 gm. of cane sugar yields 1.05 gm. of invert sugar) and mix. Transfer to a bottle, add a few drops of xylene or toluene, shake well and stopper tightly. The stock solution made in either way keeps indefinitely.

(*b*) A solution containing 1 mg. of sugar per 10 cc (5 cc of the stock solution diluted to 500 cc.).

(*c*) A solution containing 2 mg. of sugar per 10 cc (5 cc of the stock solution diluted to 250 cc).

The diluted solution should be preserved with a little added toluene or xylene; it is probably better not to depend on such diluted solutions to keep for more than a month.

**Description of the Methods.** Using an accurately calibrated pipette obtain 0.2 cc of blood from the finger tip or lobe of the ear. Deliver the blood into a centrifuge tube containing 3.8 cc of water. Rinse out the blood in the pipette by drawing the water up and allowing it to run out several times, and finally blowing out thoroughly. The total volume in the centrifuge tube should now be 4 cc (3.8 cc of water and 0.2 cc of blood). Stir with a very thin glass rod to ensure thorough laking of the blood. Add 0.5 cc of the 10 per cent solution of sodium tungstate and stir. Add 0.5 cc of two-thirds normal sulphuric acid. Stopper the tube tightly and shake vigorously. Remove the stopper and centrifugalize. Pipette off 3 cc of the clear solution and transfer to a Folin blood-sugar test tube. Add 2 cc of standard sugar solution containing respectively 0.2 and 0.4 mg. of dextrose to two similar tubes. Add 1 cc of water to each standard to bring the total volume to 3 cc or equivalent to that of the unknown. Add 2 cc of the alkaline copper solution to each tube.

This makes the total volume 5 cc as compared with 4 cc in the original Folin method. It is therefore necessary to use tubes having a slightly larger bulb, so that when containing 5 cc the surface of the solution comes within the constricted portion of the tube. They may be made from ordinary test tubes, as described by Folin, or from the regular blood-sugar tubes by blowing out the bulb slightly. In addition the tubes should be marked at 12.5 cc and 25 cc.

Place the tubes in a boiling water bath and heat for six minutes. Transfer to a cold water bath and let cool, without shaking, for two or three minutes. Add 2 cc of the molybdate phosphate solution to each tube. Let stand at least two minutes until all of the cuprous oxide is dissolved. Dilute the unknown with water to the 12.5 cc mark and the standard tubes to the 25 cc mark. Close with rubber stoppers, mix and transfer to colorimeter cups. Make the comparison in the usual way, using the standard that comes within the closest range of the unknown. Avoid bubbles which tend to accumulate on the bottom of the plungers. Before comparing the unknown of course the standard should be read against itself to check the adjustment of the colorimeter.

In making the calculation it should be noted that the unknown is diluted to 12.5 cc while the standards are diluted to 25 cc. The standards used are therefore equivalent to 0.1 and 0.2 mg. of dextrose respectively instead of 0.2 and 0.4 mg., as in the original Folin method. These standards cover the range of blood sugars between about 60 and 220 mg. per 100 cc. For higher concentrations the unknown should also be diluted to 25 cc and compared against the stronger standard, in which case the standard has the same value as in the orginal Folin method, namely, 0.4 mg. of dextrose. By roughly comparing the unknown with the standards just before dilution one can determine whether the unknown should be diluted to 12.5 cc or 25 cc to come within the range of the standards.

The 3 cc of solution taken for analysis represents three-fifths of 0.2 cc or 0.12 cc of whole blood. Therefore the calculation is made as follows, remembering, however, to use the proper value for the standard depending upon whether the unknown is diluted to 12.5 cc or 25 cc:

$$\frac{\text{Reading of standard}}{\text{Reading of unknown}} \times \frac{\text{mg. dextrose in standard}}{1} \times \frac{100}{0.12} = \text{mg. dextrose in 100 cc blood}$$

**Discussion.** The method described above is identical in principle and very similar in technic to the method of Folin and Wu. In fact, we have attempted to adhere as closely as possible to the original method and the variations are due to the smaller volume of blood taken for analysis. Before introducing these changes it was necessary first to check their effect on the accuracy of the determination.

In the precipitation of the blood proteins different proportions of blood and reagents are used. The original proportions might possibly be preserved—that is, 0.2 cc of blood, 1.4 cc of water, 0.2 cc of tungstate and 0.2 cc of acid, giving a total volume of 2 cc, from which after centrifugalizing difficulty is encountered in obtaining even 1 cc of solution for analysis To ensure an adequate volume of liquid for examination we have increased the total volume to 5 cc, which, after centrifugalizing, yields 3 cc for analysis. In doing so we have preserved the original concentration of tungstate, but at the same time the total amount in the portion taken for analysis is slightly increased. That this slight increase in tungstate is not sufficient to effect the determination is indicated by the following table:

TABLE I—RESULTS OBTAINED WITH FOLIN-WU METHOD AND MICRO-MODIFICATION.

| Blood No. | Folin-Wu, per cent. | Micro-modification, per cent. |
|---|---|---|
| 1 | 0.0588 | 0.0570 |
| 2 | 0.0843 | 0.0850 |
| 3 | 0.0913 | 0.0925 |
| 4 | 0.0950 | 0.0960 |
| 5 | 0.0970 | 0.1090 |
| 6 | 0.1040 | 0.0985 |
| 7 | 0.1110 | 0.1140 |
| 8 | 0.1140 | 0.1170 |
| 9 | 0.2100 | 0.2000 |
| 10 | 0.2200 | 0.2240 |

The table also shows the degree of accuracy of the method as compared to that of Folin and Wu.

It was also necessary to establish the reducing value of the copper solution to the smaller amounts of sugar encountered in using 0.2 cc of blood and also to the change in concentration due to taking 3 cc instead of 2 cc of filtrate for analysis. Working with pure sugar solutions and using the technic of the micro-modification we have covered the range of blood sugars ordinarily encountered. Very good results were obtained, as shown by the following table:

TABLE II.—RESULTS OBTAINED USING PURE DEXTROSE SOLUTIONS OF CONCENTRATIONS ORDINARILY ENCOUNTERED IN THE EMPLOYMENT OF THE MICRO-MODIFICATION.

| Theoretical blood sugar, per cent. | Amount of dextrose in 3 cc of filtrate, mg. | Amount of dextrose by analysis, mg. |
|---|---|---|
| 0.06 | 0.072 | 0.075 |
| 0.07 | 0.084 | 0.083 |
| 0.08 | 0.096 | 0.098 |
| 0.09 | 0.108 | 0.106 |
| 0.1 | 0.12 | 0.125 |
| 0.11 | 0.132 | 0.138 |
| 0.12 | 0.144 | 0.149 |
| 0.13 | 0.156 | 0.152 |
| 0.14 | 0.168 | 0.170 |
| 0.15 | 0.180 | 0.176 |
| 0.20 | 0.240 | 0.242 |
| 0.25 | 0.300 | 0.305 |
| 0.30 | 0.360 | 0.364 |
| 0.35 | 0.420 | 0.410 |

Before using the modification suggested the paper of Folin and Wu should be carefully read, all precautions noted and finally a series of duplicate determinations made to check the accuracy of the micro-modification with the orginial method.

In case a number of determinations are desired on patients in the hospital the centrifuge tubes, containing 3.8 cc of water, may be prepared in the laboratory and carried to the bedside, the various bloods taken, transferred to the tubes and finally returned to the laboratory for analyses.

**Conclusions.** 1. A micro-modification of the method of Folin and Wu is described for the determination of blood sugar in 0.2 cc of blood, which may be obtained from the finger or lobe of the ear.

2. We have found the method of value for clinical purposes in supplementing the original method when blood cannot be obtained from a vein.

---

# THE ROLE OF CONCENTRATED CEREAL-MILK MIXTURES IN EARLY INFANCY.

By Gaylord W. Graves, M.D.,
NEW YORK.

(From the Department of Diseases of Children of Columbia University.)

The activity of diastatic and amylolytic ferments in the digestive functions of the newborn has long been recognized. It has been noted, moreover, that even in the first days of life there is a rapid increase in power to digest starch. This capacity is highly developed by the eighth month and is said to undergo little increase during the succeeding two years.[1] Due attention has been paid to the infant's apparent need for cereal to supplement milk during the last months of the first year, and the addition of barley or other similar finely divided cereal to the formula by the third month, at least, has been customary. Foremost reasons for this practice have been the protective colloid action of the cereal, notably barley, in ensuring advantageous subdivision of casein curds; and the comparatively slow breaking down and absorption of starch in a manner not conducive to the fermentation so noticeable at times when carbohydrate of less complex molecular structure is employed exclusively.

Notwithstanding these facts the administration to very young infants of gruel mixtures of almost solid consistency, containing 10 to 15 per cent starch, is a comparatively new departure and

---

[1] Morse and Talbot: Diseases of Nutrition and Infant-feeding, Chapter III.